

Claims

1. A method of identifying active bioremediation pathways at a site comprising:

- a) contacting the microbial flora at the site with a solid support loaded or coated with a substrate that comprises an isotope;
- b) incubating said solid support for a period of time sufficient to establish a biofilm on said solid support;
- c) identifying biomarkers into which isotopes have been incorporated; and
- d) correlating the biomarkers with particular microbes or components of a bioremediation pathway.

2. The method according to claim 1, wherein said biomarkers are selected from the group consisting of lipids, nucleic acids, proteins, and carbohydrates.

3. The method according to claim 1, wherein said biomarkers are respiratory quinones, diglycerides, sterols, intact phospholipids, poly beta-hydroxyalkonates, archaeol and caldarchaeols, ornithine lipids, sphingolipids, carotenoids, glycerides, glycolipids, gangliosides, eicosanoids, hopanes, isoprenoids, terpenes, fatty acids, fatty alcohols, waxes, fatty aldehydes, proteolipids or lysolipids

4. The method according to claim 2, wherein said biomarkers are characteristic of a subset of microbial organisms.

5. The method according to claim 3, wherein said biomarkers are characteristic of a subset of microbial organisms.

6. The method according to claim 1, wherein said isotope is  $^2\text{H}$ ,  $^{13}\text{C}$ ,  $^{15}\text{N}$ , an isotope as set forth in Table 1 or a naturally occurring isotope.

7. The method according to claim 1, wherein said site is selected from the group consisting of: liquid samples; environmental samples; clinical samples; veterinary samples;

agricultural samples; food samples; gaseous samples; liquid samples; solid environmental matrices or samples; “down-well” groundwater, various water bodies, microcosms, reactors, pipes, tanks, subsurface sites, and vapor fields.

8. The method according to claim 1, wherein the biomarkers are identified by one or more of the following methods: pyrolysis, optionally with *in situ* derivatization and isotope ratio mass spectrometry; phospholipids fatty acid (PFLA) analysis, polymerase chain reaction denaturing gradient gel electrophoresis (PCR-DGGE),, expanded signature lipid biomarker analysis (SLB), terminal restriction fragment length polymorphism (T-RFLP) analysis of 16S rDNA or specific genes, high performance/atmospheric pressure chemical ionization/tandem mass spectrometry (HPLC/APCI/MS/MS) or liquid chromatography/tandem mass spectrometry (LC/MS/MS).

9. A method of identifying the microbial flora at a site comprising:

- a) contacting the microbial flora at the site with a solid support loaded or coated with a substrate that comprises an isotope;
- b) incubating said solid support for a period of time sufficient to establish a biofilm on said solid support;
- c) identifying biomarkers into which isotopes have been incorporated; and
- d) correlating the biomarkers with particular microbes or components of a bioremediation pathway.

10. The method according to claim 9, wherein said biomarkers are selected from the group consisting of lipids, nucleic acids, proteins, and carbohydrates.

11. The method according to claim 9, wherein said biomarkers are respiratory quinones, diglycerides, sterols, intact phospholipids, poly beta-hydroxyalkonates, archaeol and caldarchaeols, ornithine lipids, sphingolipids, carotenoids, glycerides, glycolipids, gangliosides, eicosanoids, hopanes, isoprenoids, terpenes, fatty acids, fatty alcohols, waxes, fatty aldehydes, proteolipids or lysolipids

12. The method according to claim 10, wherein said biomarkers are characteristic of a subset or microbial organisms.

13. The method according to claim 11, wherein said biomarkers are characteristic of a subset or microbial organisms.

14. The method according to claim 9, wherein said isotope is  $^2\text{H}$ ,  $^{13}\text{C}$ ,  $^{15}\text{N}$ , an isotope as set forth in Table 1 or a naturally occurring isotope.

15. The method according to claim 9 wherein said site is selected from the group consisting of: liquid samples; environmental samples; clinical samples; veterinary samples; agricultural samples; food samples; gaseous samples; liquid samples; solid environmental matrices or samples; "down-well" groundwater, various water bodies, microcosms, reactors, pipes, tanks, subsurface sites, and vapor fields.

16. The method according to claim 9 wherein the biomarkers are identified by one or more of the following methods: pyrolysis, optionally with *in situ* derivatization and isotope ratio mass spectrometry; phospholipids fatty acid (PFLA) analysis, polymerase chain reaction denaturing gradient gel electrophoresis (PCR-DGGE),, expanded signature lipid biomarker analysis (SLB), terminal restriction fragment length polymorphism (T-RFLP) analysis of 16S rDNA or specific genes, high performance/atmospheric pressure chemical ionization/tandem mass spectrometry (HPLC/APCI/MS/MS) or liquid chromatography/tandem mass spectrometry (LC/MS/MS).